PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

A Method for the Investigation of Fossils by Serial Sections

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VIII. *A Method for the Investigation of Fossils by Serial Sections.*

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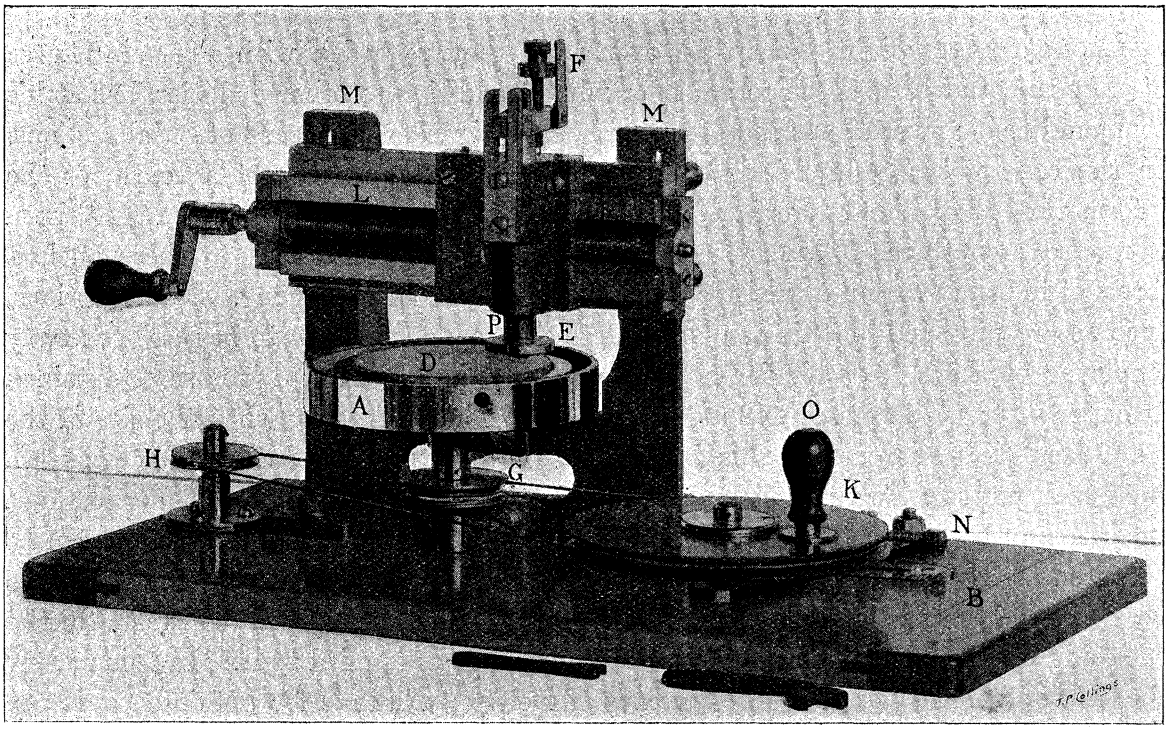
Palæontologists might perhaps be excused if they regarded with a certain degree of envy the student of recent organisms, who in addition to other facilities, has at his command that powerful method of research, investigation by means of serial sections. In the case of many important fossils not a half, frequently not a tenth, of the information they could afford, if accessible to this method, has at present been extracted from them. This is the more to be regretted since it is to these ancient forms of life that we must turn for the surest guidance in phylogeny. Serial sections, such as are provided by thin transparent slices are never likely to be very generally employed by the Palæontologist, since they cannot by any known process be obtained at closer intervals than one millimetre or more. Thus it is only when the size of the fossil is so great as to render this interval negligible, that serial thin slices can serve their real purpose. Fortunately the same objection does not apply to opaque sections, such as may be obtained by grinding away the substance of an object; these can be prepared at any desired degree of proximity. Consequently whenever the substance of a fossil, viewed by reflected light, offers a sufficient optical contrast to the enclosing matrix, the method of serial sections may be applied to it. In this matter the Palæontologist has, therefore, no cause for envy; a means for obtaining a deeper insight into the objects of his study lies ready to hand. The only difficulties are the practical; the object to be studied after having been ground through a known interval, must be removed from the grinding apparatus for examination, and its form recorded by drawing or photography; on replacing it in the apparatus to be ground through a second interval, it must be attached in such a manner that its polished face is returned to precisely the same plane that it occupied before removal. While attempting to devise a machine to accomplish this, I applied for assistance to my friend and colleague Mr. Jervis-Smith, Reader of Mechanics in the University, who at once interested himself in the matter in the kindest way, and solved the problem by designing the apparatus which is represented in figs. 1 and 2, and described by Mr. Jervis-Smith as follows:—"Two planed pillars MM, fig. 1, forming parts of one casting, are supported on a base B. The pillars carry a transverse slide and

carriage moving on it, driven by a screw and handle; the transverse slide may be adjusted to any required height from the grinding disc D, by means of screws attached to its back. A plate or chuck E carries the specimen; this plate is attached to a vertical rod, which can be raised or lowered by means of a micrometer screw F, the end of the screw acting as a stop, when the desired amount of the specimen has been removed by the grinding disc D. The wash from the disc is caught in a circular trough A, to which a tube is attached to take away the liquid used in grinding.

"The disc D (the diameter of which is 4 inches) is rotated by means of a cord

Fig. 1.

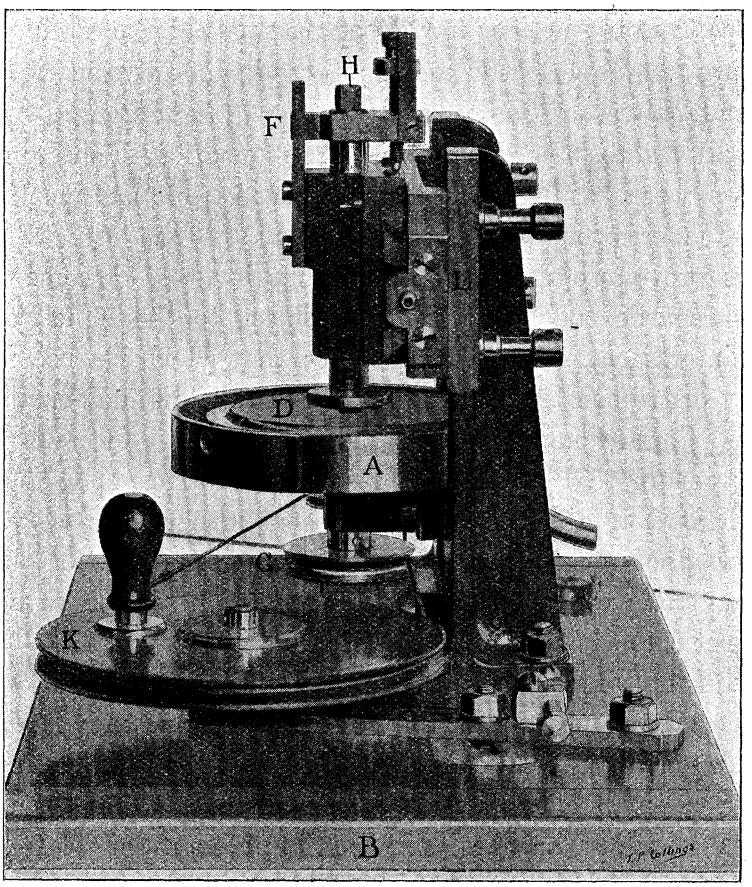


passing partly round the wheels K, H, and entirely round the wheel G. By means of this arrangement of the pulley-wheels the tension of the driving band or cord is maintained between the wheels K and H without the axle of the central wheel being subjected to injurious pressure. The grinding disc is of thick plate-glass, carried on a gun-metal disc attached to the axle of the central wheel. In the process of grinding the disc D is fed with water containing the finest emery flour in suspension. The axle of the wheel K, which is rotated by the handle O, is placed at a small angle with respect to the axle of the wheel H; this prevents the driving cord from rubbing against itself. The slack of the driving cord is taken up by means of the screw N."

It may be added that the object is restored to its original position, after removal

from the machine, by the following device. The chuck E, which carries the object, is provided with a cylindrical neck which fits into the hollow cylinder P, against which it is stopped by a flange. A steel rod passes through the axis of P and screws into the neck of the chuck, it is provided with a hexagonal head H, which stops it against the top of P; on turning this head by a spanner the chuck is tightened against P, and thus can always be returned to the same position. On bringing the chuck with its attached object over the grinding disc the polished face of the latter will be

Fig. 2.



found, if no other adjustments have been made, to rest on the surface of the grinding disc as it did before its removal.

A machine was constructed for me from Mr. Jervis Smith's drawings by Mr. R. W. Munro, of King's Cross Road. Its cost was defrayed by a grant from the Government Fund at the disposal of the Royal Society. The machine has been in almost constant use in my laboratory for the past three years, and its performance has more than satisfied my expectations. It can be trusted to give a series of sections separated by a constant interval of 0·025 millim. No attempt has been made to

obtain closer sections than this; but if greater proximity were required there can be no doubt it could be obtained.

As each section is prepared it is drawn with the aid of a camera lucida, or photographed under a microscope. Photography has many advantages over drawing, particularly as affording a record which may be trusted in questions of dispute, and this is all the more necessary, as the fossil is necessarily destroyed in the process of grinding. Photography is, however, expensive, and much more laborious than tracing with a camera.

From a series of sections the fossil may be reconstructed by any of the usual methods; cutting out from thin sheets of bees-wax and superposing gives excellent results; in this way reconstructions have been prepared of several important fossils of which the structure was imperfectly known; these include two species of a Graptolite (Monograptus), an Ophiurid of Ludlow age (*Lapworthura miltoni*), (*Ophiura egertoni*) from the Lias, and *Palæospondylus gunni*, a problematical Fish from the Old Red Sandstone. These were exhibited at the British Association meeting in Glasgow (1901), and at a Conversazione of the Royal Society (1902).

It may perhaps prove useful if a short account is added of some of the details of the method. As regards the affixing of the object to the chuck of the grinding machine, this is simply accomplished by cement; that which we use is a black wax, such as is used by instrument-makers, which melts at a low temperature, and secures the object firmly. Care should be taken that the cement runs well round the edges of the object; to insure this a strip of glass (a microscope-slide answers perfectly well) should be heated in the flame of a Bunsen burner and used to push the wax towards the object already cemented on; the wax melts in front of the glass, and on reaching the object runs up its sides by surface tension. There is not the slightest danger of the object coming loose after this treatment.

A matter of the first importance is to provide some fixed marks or registration lines, by which the relative position of the successive sections may be determined. For this purpose the fragment of rock containing the fossil should be ground into the form of a parallelopipedon, small enough for the sides or at least one corner to fall within the field of the microscope, or the limits of the photographic plate. The grinding machine may be used for this purpose, its plate glass disc being replaced by one of fine emery; the fragment of rock is cemented on to the chuck, fossil side upwards, and ground away to a plane surface; it is then detached by warming the chuck in the flame of a Bunsen, and cemented on to a brass plate specially made for the purpose, this is "L" shaped in transverse section and presents two plane surfaces at right angles to each other; the ground surface of the object is attached to one of these, and the other is cemented to the chuck. On grinding down a second face is now obtained at right angles to the first. By repeating the operation with the object readjusted, a sufficient number of times, a parallelopipedon, exact enough for all practical purposes, is obtained. In cases where the object is too large for this

treatment, three holes may be drilled in it at right angles to its surface by a hard steel bit, and into these thin cylinders of graphite, such as are used in black-lead pencils, may be inserted and fixed by some kind of cement.

After this preparation the fossil will require to be thoroughly cleaned, which is best effected by placing it in benzole, and leaving it to stand till all trace of the dark cement has been removed. It is then dried by warming over a flame.

It is now necessary completely to conceal the exposed surface of the fossil in a layer of some opaque substance, so that both may be afterwards ground away together. Unless this is done, sections with clear outlines will not be obtained, for the ground surface will be confused with the natural surface of the fossil. After various trials we have adopted red sealing-wax for this purpose; it must be of the best quality, inferior kinds are worse than useless. It grinds away smoothly and readily, but is liable to form "smears," unless excess of water is kept flowing over the grinding wheel while rotating. It accurately adapts itself to all inequalities of the object, the outlines of which it defines in the sharpest possible manner. Its melting point is above that of the cement used for attachment, so that it can be applied to the object before this is affixed to the chuck.

The object should be warmed and the wax melted in the ordinary way dropped upon it, a strip of glass, heated in a Bunsen flame, should then be applied to smooth out the melted layer and to reduce it to the least possible thickness, consistent with complete concealment of the object. A thin layer of wax should also be run round the sides of the parallelopipedon when the object has been reduced to this form, so as to insure the sharp definition of their outlines when exposed by grinding.

The object is now ready for grinding, it is attached to the chuck, which is then screwed into its support and brought over the grinding wheel. At first the sealing-wax covering alone is ground away, and the chuck must be removed from the machine and the ground surface examined under a microscope at regular intervals, as each fortieth of a millimetre is removed, until the first section of the fossil is exposed. This and all succeeding sections must then be drawn or photographed.

To increase the definition of the section, the ground surface should be covered with a drop of water, to which glycerine may be added to retard evaporation, and a cover-glass superposed.

In photographing, Welsbach burners are used for illumination; the time of exposure will vary with the optical character of the object and the matrix in which it is embedded, as well as, of course, with the magnification of the image and the nature of the sensitive plate. There is no advantage in using slow plates, and we prefer the fastest to be obtained. With an EDWARD'S isochromatic instantaneous plate, and a magnification of 20 diameters, a good photograph may be obtained in 5 minutes, when there is great contrast in actinic effect between the object and the matrix; with less contrast the exposure may be increased up to half an hour. If longer is required the case is hopeless. Our exposures have usually taken from 10 to 20 minutes.

The precaution should always be taken not to grind a fresh section till the photograph of the previous one has been developed and found satisfactory.

The negatives should be numbered in order as they are obtained, and stored with a note of the intervals they represent and the magnification employed.

As regards printing from the negatives, definition is all important, and this is best obtained with silver paper, which should be "squeegeed" down.

From the paper prints the outlines of the sections may be traced on to transparent paper; if they do not show through clearly enough, the prints and paper may be placed over a sheet of glass through which the light is directed, or the outlines may be reinforced by tracing an inked line round them, or if this fails, by substituting a 5-per-cent. solution of nitric acid for ink; this destroys the colour of the print and enables us to margin the outlines of the section with a white band.

In reconstructing the form of the fossil in wax, the usual methods are employed; a thin sheet of bees-wax of known thickness is placed over the tracing, and the outlines of the section followed by a finely pointed scalpel. The tracing paper should rest on a hard surface during this operation; a sheet of glass answers every purpose. By superposing the sections cut out in wax, in their natural order, and in their true relative positions, which are determined by the registration lines already referred to, a precise representation of the original object is obtained.

Most of our sections have been taken at intervals of one-fortieth of a millimetre, and photographed under a magnification of 20 diameters, hence the thickness of the wax sheets used in reconstruction has been approximately 0·5 millim. This is a very convenient thickness, as it allows the outline of the tracing to be easily seen through.

We have tried various methods of causing the successive wax sections to cohere; the simplest is to pass a hot needle through the sheet to be attached into those beneath it, the melted wax fills the interspace by surface tension, and by instantly applying the pressure of a finger the sheets are brought into more intimate contact. A much more elegant plan, since it leaves no unsightly marks on the model, is to fit a thin shred of wax into the angles where the edge of one sheet meets the surface of another, *i.e.*, along the contour lines, and then to run a hot needle along this, applying pressure immediately afterwards. The application of pressure is essential if true proportions are to be preserved in the model, the thickness of which must always be measured not merely by the sum of the thickness of the wax sheets, but by this *plus* the sum of the thickness of the interspaces between them. With every precaution these cannot wholly be annihilated, and consequently true proportions cannot be exactly maintained.

As a detail apparently trifling, but of great importance, both for insuring exactitude and facilitating the work of superposition, it may be added that in cases where the section presents isolated parts, these should be brought into connection with the rest by leaving strips of wax between them; economy in the use of these

ties is misplaced, at least three should connect every isolated part with various points of the remainder; after the sheet is superposed and attached, the ties can easily be removed by a hot needle. Much of the success of preparing a good model depends on keeping all the parts of the sections well tied together.

No doubt this is all well known to professional biologists, but we have found information very difficult to obtain, and have had to discover it for ourselves by the method of trial and error.

In building up a wax model some support is frequently required by the part already completed before further sections can be added. A very simple and efficient means of providing this is as follows: A quantity of Swedish filter-paper is placed in a basin of water and beaten up into a pulp; the pulp is then placed on the surface of the model to be supported; to prevent it entering too deeply into the recesses of the model, the latter may be first covered with very thin tissue-paper. When the model is completely buried in the pulp, excess of water may be removed by blotting-paper, the rest is allowed to pass off by evaporation; about 24 hours are required to complete the process of drying.

In this way very light, soft and consistent supports are obtained. The wax models may be permanently mounted by the same method.

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Fig. 1.

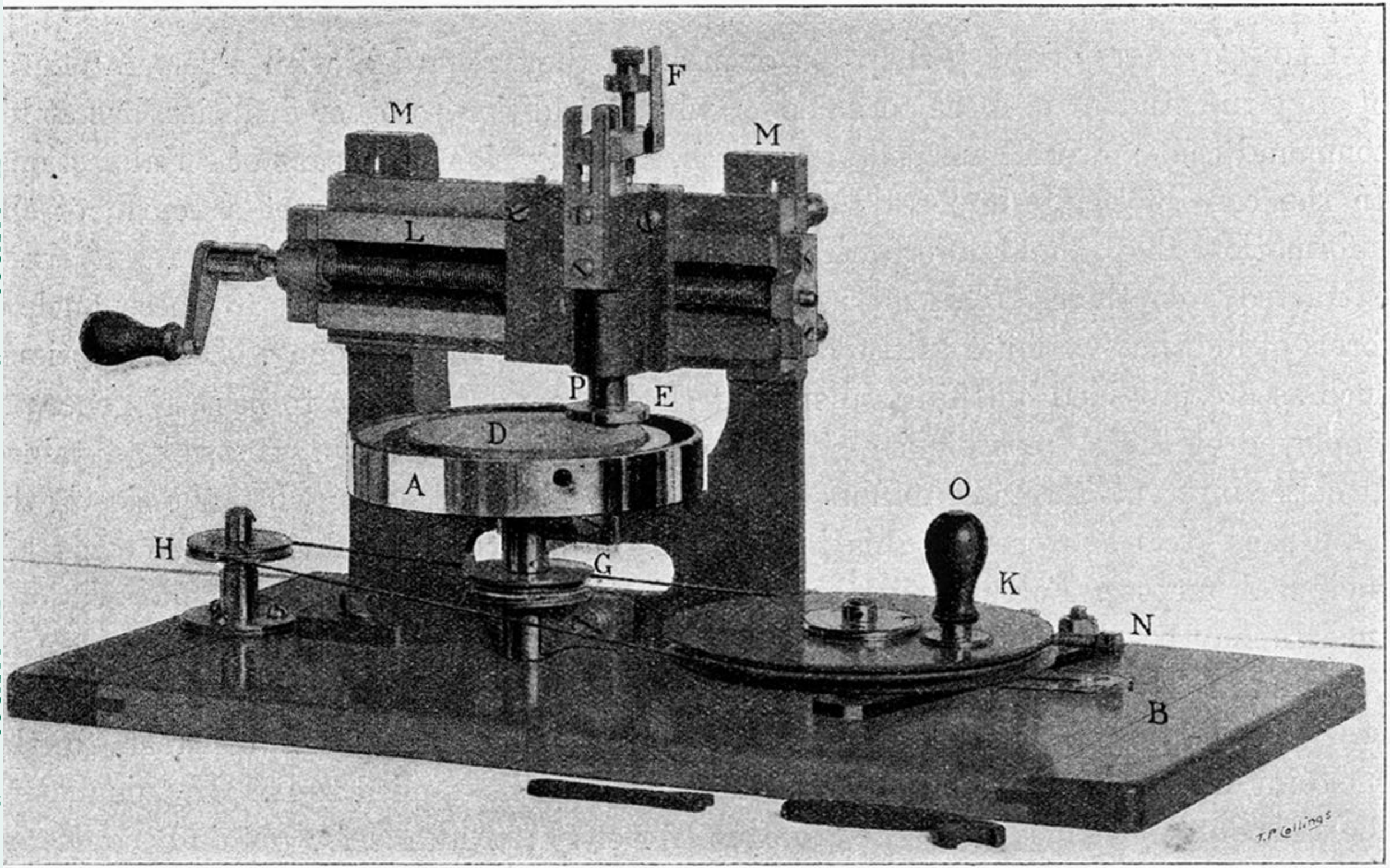

Fig. 2.